

Listing of Claims

This listing of claims will replace all prior versions and listings of claims in the application.

1. (Currently amended) ~~A method of analyzing cells disposed in media within a vessel by analyzing constituents extracted from or secreted into the media by the cells, the method comprising the steps of:~~

- ~~a) providing a vessel having an original volume of media about the cells;~~
- ~~b) reducing the original volume of media about at least a portion of the cells in the vessel to define a reduced volume of media;~~
- ~~c) analyzing a constituent related to the cells within the reduced volume of media in the vessel; and,~~

~~d) increasing the reduced volume of media about the cells in the vessel to substantially the original volume~~ determining a cell constituent extracted from or secreted into a volume of medium in a vessel by a sample comprising cells, the method comprising:

(a) placing said sample in a vessel in an apparatus comprising:

the vessel, including a chamber containing an original volume of medium and, within the chamber, a surface to hold or contain the sample;

a moveable barrier which alters the volume of the medium in contact with the sample; and

a sensor in sensing contact with the medium;

(b) moving the barrier to reduce the volume of the chamber and thereby the medium, forming a reduced-volume measuring chamber in contact with the sample;

(c) determining a cell constituent in the medium in the reduced volume measuring chamber with the sensor; and

(d) moving the barrier to increase the reduced volume of medium about the cells to substantially the original volume.

2. (Previously presented) The method of claim 1, further comprising the step of repeating steps b) and c).

3. (Original) The method of claim 1, further comprising the steps of:
determining a first concentration of the constituent; and
determining a second concentration of the constituent at a predetermined time interval from determining the first concentration.

4. (Original) The method of claim 3 further comprising the step of calculating a flux rate of the constituent based on the first concentration and the second concentration.

5. (Original) The method of claim 1 wherein the reduced volume is in a range of about 5-50% of the original volume.

6. (Original) The method of claim 5 wherein the reduced volume is in a range of about 5-20% of the original volume.

7. (Original) The method of claim 1 wherein the reduced volume is less than about 5% of the original volume.

8. (Original) The method of claim 1 wherein the cells comprise a cell selected from the group consisting of bacteria, fungus, yeast, a prokaryotic cell, a eukaryotic cell, an animal cell, a human cell, and an immortal cell.

9. (Currently amended) The method of claim 1 wherein at least a portion of the cells are attached to said surface of ~~the vessel~~ said chamber.

10. (Original) The method of claim 1 wherein at least a portion of the cells are suspended in the media.

11. (Original) The method of claim 1 wherein at least a portion of the cells comprise living tissue.

12. (Original) The method of claim 1 wherein the constituent comprises a material selected from the group consisting of a dissolved gas, an ion, a protein, a substrate, a salt, and a mineral.

13. (Original) The method of claim 12 wherein the dissolved gas is selected from the group consisting of O₂, CO₂, and NH₃.

14.-16. (Cancelled)

17. (Original) The method of claim 1 wherein the constituent comprises a material extracted from the media by at least a portion of the cells.

18. (Original) The method of claim 1 wherein the constituent comprises a material secreted into the media by at least a portion of the cells.

19. (Currently amended) The method of claim 1 wherein ~~determining~~analyzing the cell constituent comprises sensing a presence of the constituent.

20. (Currently amended) The method of claim 1 wherein ~~determining~~analyzing the cell constituent comprises sensing a concentration of the constituent.

21. (Currently amended) The method of claim 1 wherein ~~determining~~analyzing the cell constituent comprises sensing a first concentration of a first constituent, sensing a second concentration of a second constituent, and determining a relationship between the first concentration and the second concentration.

22. (Currently amended) The method of claim 1 wherein ~~determining~~analyzing the cell constituent comprises sensing a rate of change of concentration of the constituent.

23. (Cancelled)

24. (Currently amended) The method of claim ~~23~~ 1 wherein the sensor comprises a sensor selected from the group consisting of a fluorescent sensor, a luminescent sensor, an ISFET sensor, a surface plasmon resonance sensor, a sensor based on an optical diffraction principle, a sensor based on a principle of Wood's anomaly, an acoustic sensor, and a microwave sensor.

25. (Currently amended) The method of claim 1 wherein ~~determining~~analyzing the cell constituent comprises determining a parameter selected from the group consisting of cell viability, cell number, cell growth rate, response to at least one of a drug, a toxin, and a chemical, detection of an entity, and internalization.

26. (Original) The method of claim 1, further comprising the step of perfusing additional media through the vessel.

27. (Original) The method of claim 1, further comprising the step of replenishing the media in the vessel.

28. (Cancelled)

29. (Currently amended) The method of claim ~~28-1~~ wherein the barrier is disposed in the vessel without causing displacement of media out of the vessel.

30. (Currently amended) The method of claim ~~28- 1~~ wherein at least a portion of the barrier comprises the sensor.

31. (Currently amended) The method of claim ~~28- 1~~ wherein ~~[[a]]~~ the sensor is disposed on a surface of said barrier in contact with said reduced volume measuring chamber.

32. (Original) The method of claim 31 wherein the sensor comprises a fluorophore.

33. (Original) The method of claim 1 wherein at least a portion of the vessel comprises a sensor.

34. (Original) The method of claim 1 comprising the additional step of altering an environment of at least a portion of the cells prior to reducing the original volume of media.

35. (Original) The method of claim 34 wherein altering the environment comprises exposing at least a portion of the cells to at least one of a drug, a chemical, and a toxin.

36. (Currently amended) The method of claim 1, further comprising the step of altering an environment of at least a portion of the cells after ~~reducing the original volume of media-step b).~~

37. (Original) The method of claim 1 comprising the additional step of covering the vessel.

38. (Original) The method of claim 1 comprising the additional step of stirring at least a portion of the original volume of media in the vessel.

39. (Original) The method of claim 1 comprising the additional step of sealing the vessel.

40.-80. (Cancelled)

81. (Previously presented) The method of claim 1 wherein the cells comprise an animal cell, a human cell, or an immortal cell.

82. (Cancelled)

83. (Previously presented) The method of claim 1 wherein said vessel comprises a well in a plate including multiple wells for holding media and cells.

84. (Previously presented) A method of analyzing cells disposed in media within a vessel by analyzing constituents extracted from or secreted into the media, the method comprising the steps of:

- (a) providing a vessel having an original volume of media about the cells;
- (b) disposing a barrier into the vessel to reduce the original volume of media about at least a portion of the cells in the vessel and to define a reduced volume of media; and
- (c) analyzing a constituent related to the cells within the reduced volume of media using a sensor in contact with the reduced volume of media.

85. (Previously presented) The method of claim 84 wherein the cells are selected from the group consisting of animal cells, human cells, and immortal cells.

86. (Previously presented) The method of claim 84 wherein the sensor is disposed on a surface of said barrier in contact with the reduced volume.

87. (Previously presented) The method of claim 84 wherein the sensor comprises a fluorophore.

88. (Previously presented) The method of claim 86 wherein the sensor comprises a fluorophore.

89. (Previously presented) The method of claim 84 comprising the additional step of :

d) increasing the reduced volume of media about the cells in said vessel to substantially the original volume.

90. (Previously presented) The method of claim 84 wherein the constituent comprises a material selected from the group consisting of a dissolved gas, an ion, a protein, a substrate, a salt, and a mineral.

91. (Previously presented) The method of claim 84 wherein the constituent comprises a dissolved gas selected from the group consisting of O₂, CO₂, and NH₃.

92. (Previously presented) The method of claim 84 wherein the sensor comprises a sensor selected from the group consisting of a fluorescent sensor, a luminescent sensor, an ISFET sensor, a surface plasmon resonance sensor, a sensor based on an optical diffraction principle, a sensor based on a principle of Wood's anomaly, an acoustic sensor, and a microwave sensor.

93. (Previously presented) The method of claim 84 wherein the barrier is disposed in the vessel without causing displacement of media out of the vessel.

94. (Previously presented) The method of claim 84 comprising the additional step of repeating steps b) and c) without removing said cells from said media.

95. (Previously presented) The method of claim 84 wherein said vessel comprises a well in a plate including multiple wells for holding media and cells.